DO TREE LEAVES PROMOTE DIGESTION OF GRASS BY CATTLE?

P.M. KENNEDY [A], *J.B. LOWRY* [B]

[A] CSIRO Livestock Industries, PO Box 5545, Rockhampton M.C. Queensland 4702.
[B] CSIRO Livestock Industries, 120 Meiers Rd., Indooroopilly, Queensland 4068.

SUMMARY

The degree of fermentation of leaves from 14 trees was ranked by relative gas pressures attained in an *in vitro* system, and four were selected for further study. Progress of fermentation with rumen microbes of fallen leaves of siris (*Albizia lebbeck*), yemane (*Gmelina arborea*), tipuana (*Tipuana tipu*), and bauhinia (*Bauhinia carronii*), by themselves or in mixtures with angleton grass (*Dichanthium aristatum*), was monitored by changes in gas pressure and recovery of cell wall as neutral detergent fibre. With the addition of N to N-poor media, responses in fibre digestion ranged from large and positive with yemane by itself or in mixtures with grass, to negative for bauhinia mixtures in which formation of artifact cell wall was observed. Positive associative (non-additive) effects on fibre digestion were recorded for 5 of the 6 leaf/media combinations for siris, yemane, and tipuana, but negative effects were seen for bauhinia mixtures. Following extraction of leaf cell solubles and fermentation of extracted leaf with grass, an increased associative effect was achieved for bauhinia when compared to the unextracted leaf:grass mixture, but no difference was observed for the other leaf types. This study indicated leaf fall from dry-season deciduous trees may provide supplementary nutrition for cattle in northern Australia, but the benefits are highly species dependent.

Keywords: associative effects, leaves, supplements, rangelands

INTRODUCTION

In pasture systems, fallen leaf from deciduous trees may make a significant contribution to nutrition of livestock (Lowry and Wilson 1999). This raises the possibility of selection of dual-purpose trees for agro-forestry that could provide nutrition for ruminants as well as timber value (Lowry and Seebeck 1997). One such tree is siris (*Albizia lebbeck),* present in northern Australian rangelands, the leaf of which, offered to cattle as a supplement to a mature grass diet, markedly increased digestible dry matter (DM) intake (Kennedy *et al*. 2002). In that study, we also suggested that when leaves of siris were co-fermented with angleton grass (*Dichanthium aristatum*), positive associative (non-additive) effects occurred.

The progress of digestion and the presence of associative effects between leaf and poor quality grass may be assessed following fermentation by rumen microbes *in vitro* as the loss of fibre or as the volume of gas produced from fermentation with rumen microbes (Wood and Manyuchi 1997). Both methods have deficiencies. Compounds insoluble in neutral detergent may be produced as complexes between protein in leaf cell wall and phenolic molecules or proanthocyanidins and hence be measured as fibre (Reed 1986; Merkel *et al*. 1999). Whether this ‘artifact’ fibre leads to depressed fermentation is uncertain, and a supplementary index of fermentation is desirable. Gas production is one such index, but includes gas from fermentation of soluble material, and its relationship to fermentation of substrate is dependent on stoichiometry, which may change with substrate, microbial inoculant, and as digestion progresses.

The present study investigated fermentation *in vitro* of fallen leaves of 14 trees with potential as fodder trees in northern Australian rangelands. Subsequently four were selected on the basis of their potential value as dual-purpose trees for timber production and providing nutrients to cattle. These species were siris, yemane (*Gmelina arborea*), tipuana (*Tipuana tipu*), and bauhinia (*Bauhinia carronii*).

MATERIALS AND METHODS

Procedures and measurements

In experiment 1, fallen leaves of siris, yemane, tipuana, bauhinia, *Albizia procera, Brachychiton acerifolium, Albizia toona, Pongamia pinnata, Carissa ovata, Albizia basaltica, Atalaya hemiglauca, Bursaria incana, Eucalyptus platyphylla,* and *Melia azedarach* were ground (1 mm screen) and fermented alone or mixed 50:50 (DM basis) with ground angleton grass in complete (GVS) media with 3 ml of rumen inoculum. On the basis of their potential as dual-purpose trees, and fermentation rating of moderate or above, the first four leaf substrates were chosen for further study.

In experiment 2, leaf samples of siris, yemane, tipuana and bauhinia were fermented alone or mixed with angleton grass in 25:75, 50:50, 75:25 and 100:0 ratios of grass:leaf, in media poor (NP) or rich (NR) in N. In experiment 3, the four leaf types were sequentially extracted with hot ethanol and boiling water, and 1 ml aqueous extract was added to angleton grass, to supply leaf cell soluble material equivalent to that contained in an equal mixture of leaf and grass, and fermented in GVS media. In addition, leaf or extracted leaf was fermented alone or with angleton grass, in order to establish if removal of soluble material affected values of associative effects. The ratio of leaf:grass was 50:50 for non-extracted leaf, and that for each extracted leaf type was calculated to provide the same content of leaf NDF as its non-extracted counterpart. For description of procedures used, see Kennedy *et al*. (2002). Formation of artifact NDF was indicated if NDF recovery increased between 0 and 6 h of fermentation.

Calculations and statistical analysis

Responses to N supplementation were calculated as the difference in NDF digestion of substrates in NR and NP media, expressed as a percentage of initial NDF content of substrate, i.e., changes in NDF digestibility at each sampling time. Responses in gas production were expressed as the percentage increase in gas pressure for fermentation in NR, compared to NP media. Associative effects were calculated within media as the difference in NDF digestibility between that measured in leaf:grass mixtures, and that expected from NDF digestion of leaf and grass when fermented alone (Kennedy *et al*. 2002), and are reported as %NDF digestibility units. Associative effects in gas production were expressed as the difference in observed gas pressure and that expected from the component leaf and grass, expressed as a percentage of observed values.

Results were subjected to analysis of variance. Effects of sampling time, media or substrate were tested against residual. When the F-test was significant ($P<0.05$), planned comparisons were made within sampling time between multiple means based on least significant differences.

Some of the results with siris were reported previously (Kennedy *et al.* 2002).

RESULTS

In experiment 1, yemane, *Pongamia*, *Brachychiton*, *Bursaria* and *Melia* leaves were relatively fermentable, with gas production of 78-85% of that from angleton grass after fermentation for 72 h. Gas evolved from siris and tipuana was about 65% of that from grass. Lower gas production (40-50% of that from grass) was seen for bauhinia, *Carissa*, *Atalaya*, and *A. basaltica*, and there was evidence of artifact NDF. *A. toona*, *A. procera*, and *E. platyphylla* also had moderate gas production, with evidence of artifact NDF for the *Albizia* spp., and all were strongly inhibitory in co-fermentation with grass.

In experiment 2, when NDF digestibility in NP and NR media was compared, it was found that N addition did not affect digestion of leaf alone for siris, tipuana and bauhinia, but for yemane there was an increase by 6 %digestibility units ($P<0.05$) after 12 h; this difference progressively disappeared by 28 h. For grass fermented alone in 4 runs, addition of N to the media lead to a significant increase ($P<0.05$) in NDF digestibility of 4.5-7 units during mid-fermentation for experiments involving siris, yemane and bauhinia, in contrast to a larger (19 units) response in the experiment involving tipuana, indicating a greater N deficiency in that rumen inoculum.

Responses to extra media N, and associative effects for the 3 grass:leaf ratios within 4 leaf types, are presented in Table 1. Associative effects for NDF digestion of 3 %digestibility units or more were generally significant ($P<0.05$), and values of 2 to 3 units were less so ($P<0.10$). Positive associative effects were seen in both NR and NP media for siris and tipuana, and in NR media for yemane. Negative effects were noted for yemane in NP media and for bauhinia in both media. Gas pressure differences were in general qualitative agreement with conclusions from NDF data. For bauhinia mixtures, between 0 and 6 h of fermentation there was an apparent increase in NDF content of the bauhinia component by 44%, and this artifact NDF disappeared linearly over the subsequent 42 h. Equipment failure resulted in the loss of the final time data for bauhinia.

In experiment 3, NDF content of leaves ranged between 28 and 52 (Table 2). During fermentation of bauhinia leaf, 30% of the artifact NDF present at 6 h had disappeared in a linear fashion by 72 h. NDF digestibility at 72 h of siris, yemane, tipuana and bauhinia respectively when leaf was fermented alone, was 27, 47, 27 and –1% and cumulative gas pressure was equivalent to 62, 83, 56 and 44% that from grass alone. The majority (80-90%) of NDF digestion from leaf was completed by 24 h, except for bauhinia. N content of grass, siris, yemane, tipuana and bauhinia was 3.2, 17.8, 16.2, 13.5, and 10.6 g/kg DM respectively; solvent extraction removed 28, 33, 30 and 31% of leaf N respectively. Associative effects were similar within leaf types when either non-extracted or extracted leaf was used, with the exception of bauhinia, in which there was a significant ($P<0.05$) increased effect when leaf was extracted. All extracts of cell solubles were slightly inhibitory ($P<0.05$) to fermentation when added to grass; the inhibition for bauhinia was greater ($P<0.05$) than for the other leaf types (Table 2).

Table 1. Effects of additional N in media (comparison of digestion in NR and NP media), and associative effects for NDF digestibility and gas production, within media at 3 times, with mixtures of leaf and grass in 75:25, 50:50 and 25:75 ratios. Units: for gas, % change in volume; for NDF, change in %digesibility.

	Effect of N						Associative effects											
							NDF						GAS					
	NDF			GAS			NP			NR			NP			NR		
Time (h)	28	48	72	28	48	72	28	48	72	28	48	72	28	48	72	28	48	72
Grass: siris																		
75:25	2	4	1	11	9	6	6	3	2	3	3	2	9	5	2	7	8	6
50:50	5	3	1	8	4	2	6	5	4	4	5	4	7	5	1	8	8	4
25:75	2	2	2	-6	-5	-4	3	5	2	1	1	3	7	4	1	1	2	2
Grass: yemane																		
75:25	12	9	10	41	17	10	-4	-2	-1	4	5	5	-13	-7	-5	7	6	6
50:50	15	7	1	26	10	4	-5	-4	-3	5	6	6	-5	-1	2	8	8	7
25:75	15	5	11	8	4	4	2	2	2	4	5	7	4	1	2	5	6	8
Grass: tipuana																		
75:25	4	8	6	31	21	6	7	5	2	2	2	1	15	17	13	6	6	4
50:50	9	10	4	20	14	5	8	6	4	3	3	4	10	12	8	2	2	0
25:75	1	2	2	13	6	5	5	5	4	3	2	1	12	13	9	8	4	3
Grass:bauhinia																		
75:25	-3	-2		-11	-7		1	-2		-2	1		14	18		-10	-45	
50:50	-3	-2		7	5		-6	-3		-7	-2		-4	11		-30	-8	
25:75	-5	-2		36	41		-10	-6		-6	-1		-26	-7		-27	-7	

Table 2. Content and digestibility of NDF in leaf after 72 h of fermentation, extraction of DM by solvents, mean associative effects of leaf and extracted leaf when mixed with grass (50:50) and fermented for 28, 48 and 72 h in GVS media, and effects of soluble extract on NDF digestibility (%units).

	Leaf NDF content (% DM)	Leaf NDF digestibility (%)	Leaf DM extracted (%)	Associative effect (NDF digestibility)			Effect of extract on NDF digestibility		
				28	48	72	28	48	72
Siris	48	27	34	2	3	3	-4	-2	-2
Yemane	28	47	51	0	2	3	-7	-2	-2
Tipuana	52	27	34	3	4	4	-3	-6	-3
Bauhinia	31	-1	42	-1 [A]	0 [A]	0 [A]	-9	-9	-2
				3 [B]	4 [B]	1 [B]			

[A] non-extracted leaf, [B] extracted leaf

DISCUSSION

Leaf material would be of especial value for cattle performance in the dry season if it were high in rumen available-N. For siris, yemane, tipuana and bauhinia, approximately 30% of leaf N was extractable as cell solubles, however this was unlikely to be sufficient to relieve N deficiency in ruminants eating poor quality hay. Thus, even when leaf comprised 75% of the mixture in experiment 2, a response to N of NDF digestion was observed with siris and tipuana. Even greater responses were observed for all mixtures of yemane with grass, and indeed additional N stimulated digestion of yemane leaf fermented alone, in contrast to the situation with the three other leaf substrates. Rosales *et al*. (1998) reported that positive associative effects seen with tropical leaves in mixture with each other varied with time and N supply. In the current experiment, a similar variable result was obtained with yemane, for which some interactions of associative effects with N content of media were evident.

Apart from supplying N to microbes, desirable properties of leaf include high initial fermentability, and promotion of positive associative effects when co-fermented with grass. As shown by the depression of grass digestion when leaf solubles were added, a high content of readily-fermentable material may not necessarily be desirable, although it needs to be recognised that this conclusion may not apply *in vivo*. Indeed, the similar fermentation patterns for mixtures of grass with extracted or non-extracted leaf, indicated that when fermentation of cell solubles proceeded *in situ*, there was no effect except when cell solubles appeared to contain anti-microbial properties, as in bauhinia.

The basis of positive associative effects is not clear, but may involve stimulation of microbial activity by increased diversity of niches and microbial populations when leaf is mixed with grass (see Forsberg *et al*. 1997). Limited data for siris and yemane leaf, for which amount of NDF digested over 72 h was similar but associative effects differed, suggest that rates of NDF digestion or soluble material may have had an influence. Diversity of rate and extent of NDF digestion in leaf relative to grass, may allow more vigorous or extended digestion of mixed substrates.

There was general agreement between NDF and gas results indicating that the latter is a good proxy for qualitative evaluation of associative effects in fibre digestion. Formation of artifact NDF, evident with bauhinia, is an undesirable property in leaf from a fodder tree, as indicated by negative associative effects in both NDF digestion and gas production observed with bauhinia in this study.

This study indicated the potential of leaf fall from trees to provide supplementary nutrients for cattle in northern Australia. However, there are important between-species differences not apparent from content of fibre and protein. On the basis of results presented here, siris and tipuana are good candidates as fodder trees, whereas for yemane, clarification is needed of role of the higher NDF digestibility and N requirements in determining associative effects. Considering that flower and pod material also can contribute to feeding value of tree by-products (Lowry and Wilson 1999) further assessment of candidate trees, in the context of further research into potential of dual-purpose trees, appears desirable.

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Email: peter.kennedy@csiro.au